

FILE 'EMBASE, MEDLINE, CAPLUS, BIOSIS' ENTERED AT 17:41:40 ON 13 APR 2005

L1 753 S CONDITIONAL? REPLICAT?  
L2 1590774 S HEAT OR (HSP OR HSE OR HSF)  
L3 0 S L1 (S) L2  
L4 2477 S TRANSCRIPTION? TARGET?  
L5 12 S L4 (S) L2  
L6 3 DUP REM L5 (9 DUPLICATES REMOVED)  
L7 478 S L4 (P) (?VIRUS OR ?VIRAL)  
L8 1 S L1 (P) L2  
L9 216 S L4 (S) (?VIRUS OR ?VIRAL)  
L10 0 S L9 AND HEAT  
L11 0 S L9 AND HEAT?  
L12 12 S L7 AND HEAT?  
L13 3 DUP REM L12 (9 DUPLICATES REMOVED)  
L14 1 S L1 (P) HEAT?  
L15 543 S ADENOVIR? (S) HEAT?  
L16 84 S ADENOVIR? (3A) (HEAT (2A) SHOCK)  
L17 10 S L16 AND PROMOTER  
L18 10 DUP REM L17 (0 DUPLICATES REMOVED)

AU Madara Jonathan; Krewet James A; Shah Maulik

SO Molecular cancer, (2005 Mar 11) 4 (1) 12. Electronic Publication:  
2005-03-11.

Journal code: 101147698. ISSN: 1476-4598.

TI Heat shock protein 72 expression allows permissive replication of  
oncolytic adenovirus dl1520 (ONYX-015) in rat glioblastoma cells.

AB In this study we have made novel observations with regards to potentiation  
of the tumoricidal activity of the oncolytic adenovirus, dl1520 (ONYX-015)  
in rat glioblastoma cell lines expressing heat shock protein 72  
(HSP72) due to permissive virus replication. ONYX-015 is a  
conditionally replicating adenovirus that is deleted for  
the E1B 55 kDa gene product whose normal function is to interact with  
cell-cycle regulatory proteins to permit virus replication. However, many  
murine and rodent cell lines are not permissive for adenovirus  
replication. Previously, it has been reported that the heat  
shock response is necessary for adenovirus replication and that induction  
of heat shock proteins is mediated by E1 region gene products.  
Therefore, we hypothesized that HSP72 expression may allow for permissive  
replication of ONYX-015 in previously non-permissive cells. Rat glioma  
cell lines 9L and RT2 were transfected with a plasmids expressing HSP72 or  
GFP. After infection with ONYX-015, no tumoricidal activity is observed  
in GFP expressing cell lines despite adequate transduction. In contrast,  
HSP72 transfected cells show cytopathic effects by 72 hours and greater  
than 75% loss of viability by 96 hours. Burst assays show active virus  
replication in the HSP72 expressing cell lines. Therefore, 9L-HSP72 and  
RT2-HSP72 are ideal models to evaluate the efficacy of ONYX-015 in an  
immunocompetent rat model. Our study has implications for creating rodent  
tumor models for pre-clinical studies with E1 region deleted  
conditionally replicating adenovirus.

AU Bateman A.; Bullough F.; Murphy S.; Emiliusen L.; Lavillette D.; Cosset  
F.- L.; Cattaneo R.; Russell S.J.; Vile R.G.

SO Cancer Research, (15 Mar 2000) Vol. 60, No. 6, pp. 1492-1497.

Refs: 20

ISSN: 0008-5472 CODEN: CNREA8

TI Fusogenic membrane glycoproteins as a novel class of genes for the local  
and immune-mediated control of tumor growth.

AB We report here the use of viral fusogenic membrane glycoproteins  
(FMGs) as a new class of therapeutic genes for the control of tumor  
growth. FMGs kill cells by fusing them into large multinucleated

syncytia, which die by sequestration of cell nuclei and subsequent nuclear fusion by a mechanism that is nonapoptotic, as assessed by multiple criteria. Direct and bystander killing of three different FMGs were at least one log more potent than that of herpes simplex virus thymidine kinase or cytosine deaminase suicide genes. Transduction of human tumor xenografts with plasmid DNA prevented tumor outgrowth in vivo, and cytotoxicity could be regulated through transcriptional targeting. Syncytial formation is accompanied by the induction of immunostimulatory heat shock proteins, and tumor-associated FMG expression in immunocompetent animals generated specific antitumor immunity.

AU Emiliusen L.; Gough M.; Bateman A.; Ahmed A.; Voellmy R.; Chester J.; Diaz R.M.; Harrington K.; Vile R.

SO Gene Therapy, (2001) Vol. 8, No. 13, pp. 987-998.

Refs: 29

ISSN: 0969-7128 CODEN: GETHEC

TI A transcriptional feedback loop for tissue-specific expression of highly cytotoxic genes which incorporates an immunostimulatory component.

AB Transcriptional targeting of cytotoxic genes is an important way to control toxicity associated with gene transfer therapies, but supposedly, tissue-specific promoters are often either very weak and/or leaky. In addition, the phenotypic leakiness of such tissue-specific promoters is dependent upon the toxicity of the gene being used. Therefore, we devised a transcriptional feedback loop to restrict gene expression of very potent genes to melanoma cells. We screened different elements of the human tyrosinase promoter to find one which gave no detectable expression in non-melanoma cells but was active in melanoma cell lines. This weak, but highly tissue specific, element (Tyr-300) was then used as the basis for a transcriptional amplification feedback loop in which a consensus heat shock element (HSE) was cloned upstream of Tyr-300. The cytotoxic gene was cloned downstream of the HSE-Tyr-300 element along with a mutated form of the heat shock factor-1 (HSF-1) transcription factor, which no longer requires cellular stress to activate its trimerisation, nuclear localisation and transcriptional activation properties. Low levels of expression from Tyr-300 initiated expression of both the cytotoxic and the HSF-1 genes in melanoma cells. Gradual build up of HSF-1 amplified expression through binding to the HSE to give levels of cytotoxicity similar to that provided by a CMV promoter. However, no leakiness was observed in multiple non-melanoma cell lines tested. In addition to amplifying low levels of weak tissue-specific expression, the use of HSF-1 also leads to activation of endogenous stress-related genes such as hsp70. Induction of these genes, in the presence of cell killing by the cytotoxic gene, is a highly immunostimulatory event which enhances the antitumour vaccination effects of direct tumour cell destruction. Having demonstrated the compatibility of the component elements in plasmid form, we incorporated the feedback loop into a hybrid LTR-modified retroviral vector and confirmed that the system can be effective in the form of a viral vector. The format of the feedback loop described here could be exploited for any tissue type in which a highly tissue-specific element can be identified but which is itself too weak to be effective therapeutically.

AU Brade A.M.; Szmitko P.; Ngo D.; Liu F.-F.; Klamut H.J.

SO Human Gene Therapy, (20 Mar 2003) Vol. 14, No. 5, pp. 447-461.

Refs: 52

ISSN: 1043-0342 CODEN: HGTHE3

TI Heat-directed tumor cell fusion.

AB In previous studies we demonstrated that a modified human HSP70b promoter (HSE.70b) directs high levels of gene expression to tumor cells after mild hyperthermia treatment in the range of 41.5-44.degree.C. This transcriptional targeting system exhibits low basal

activity at 37.degree.C, is highly induced (950-fold) after mild heat treatment (43.degree.C/30 min), and returns to basal activity levels within 12-24 hours of activation. Here we describe heat-directed targeting of an activated form of the Gibbon ape leukemia virus env protein (GALV FMG) to tumor cells. GALV FMG mediates cell-cell fusion, and when expressed in tumor cells can produce bystander effects of up to 1:200. Transient transfection of a HSE70b.GALV FMG minigene caused extensive syncytia formation in HeLa and HT-1080 cells following mild heat treatment (44.degree.C/30 min). Stable transfection into HT-1080 cells produced a cell line (HG5) that exhibits massive syncytia formation and a 60% reduction in viability relative to a vector-only control (CI1) following heat treatment in vitro. Mild hyperthermia also resulted in syncytia formation, necrosis, and complete macroscopic regression of HG5 xenograft tumors grown in the footpads of mice with severe combined immunodeficiency disorders (SCID). Median survival increased from 12.5 (in heated CI1 controls) to 52 days after a single heat treatment. Heat-directed tumor cell fusion may prove to be a highly beneficial adjunct to existing cancer treatment strategies that take advantage of the synergistic interaction between mild hyperthermia and radiation or chemotherapeutic drugs.

- AU Borrelli, Michael J.; Schoenherr, Diane M.; Wong, Alden; Bernock, Laura J.; Corry, Peter M.
- SO Cancer Research (2001), 61(3), 1113-1121  
CODEN: CNREA8; ISSN: 0008-5472
- TI Heat-activated transgene expression from adenovirus vectors infected into human prostate cancer cells
- AB Replication-deficient adenovirus expression vectors were used to introduce a recombinant DNA construct contg. enhanced green fluorescent protein (EGFP) under control of a truncated, human heat shock promoter into human prostate cancer cells growing either exponentially or in plateau phase. This was done to measure controlled, heat shock-induced EGFP expression under conditions relevant to treating human cancers with heat-activated gene therapy. Both the temporal duration and magnitude of EGFP expression increased proportionately with stronger heat shocks (time at temp.) up to max. values that were induced by 4 h at 41.0.degree. or 2 h at 42.0.degree.. Longer heat shocks at either temp. yielded no addnl. EGFP expression and ultimately reduced it. Maximal EGFP expression was induced in exponential cultures by heat shocks delivered 12-24 h after virus infection. Induction at progressively later postinfection times induced increasingly lower, peak EGFP expression. Maximal EGFP expression could not be induced until 48 h after infection of plateau phase cultures but could still be induced 180 h after virus infection. However, peak EGFP levels in plateau cultures were approx. 25-50% of those obsd. in identically induced exponential cultures. Ostensibly, the differences in expression from the heat shock promoter obsd. in exponential and plateau cultures were attributable to cell division dilg. the vector within exponential cultures and the lower metabolic activity in serum-starved plateau cultures. For all exptl. conditions, EGFP expression induced from the heat shock promoter was comparable with or higher than that from the constitutively active cytomegalovirus promoter over any 24-h period. The exptl. results demonstrated that EGFP expression from the heat shock promoter was controllable in both exponential and plateau phase cultures and support the plausibility of using controlled heat shock activation of this promoter as a means of regulating both the spatial and temporal expression of therapeutic DNA constructs within human tissues. The ability to localize and regulate expression from the heat shock promoter may prove particularly advantageous for many cancer applications, esp. if the therapeutic products are highly toxic, e.g., proteotoxins or cytokines. However, the results of this study suggest that differential growth conditions within tumors could markedly affect

the expression of recombinant DNA under control of both inducible and constitutive promoters. Consequently, inducing schemes may need to be spatially adjusted to obtain the desired therapeutic results in all tumor domains using heat-activated gene therapy.

IN Gamerman, Gary Eric  
SO U.S. Pat. Appl. Publ., 24 pp.  
CODEN: USXXCO  
TI Vectors having replication, immunogenicity and/or pathogenicity under stress promoter regulation and use thereof  
AB The present invention relates to modified vectors, e.g. plasmids, viruses or microorganisms such as yeast or bacteria, wherein the replication, immunogenicity and/or pathogenicity is placed under the control of at least one stress gene regulating element. It is an object of the invention to utilize stress inducible regulatory elements, e.g., heat shock regulatory elements to regulate the replication, immunogenicity and/or pathogenicity of an autonomously replicating vector which in its wild-form does not replicate and/or exhibit pathogenicity or immunogenicity under heat shock control. It is another object of the invention to provide adenoviral vectors wherein the replication, immunogenicity and/or pathogenicity is modulated by placing a gene, e.g. an adenoviral replication gene, under the control of at least one heat shock regulatory sequence. In preferred embodiments, these modified vectors are used for gene therapy, in vaccines, or for functional genomic screening.

Ref #	Hits	Search Query	DBs	Default Operator	Plurals	Time Stamp
S1	15	richard near2 voellmy.in.	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 16:53
S2	0	10/731,961	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 16:49
S3	13759	heat same (viral or virus)	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 16:53
S4	1631	S3 same replication	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 16:53
S5	24218	(virus OR viral) near3 replication	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 16:54
S6	52	S5 with heat	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:08
S7	728	conditional\$ near2 replicat\$	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:09
S8	306	S7 same (virus OR viral)	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:10
S9	388	S7 with (adenovir\$ OR retrovir\$ OR herpesvir\$)	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:14
S10	0	S9 with heat	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:11
S11	0	S9 same heat	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:12
S12	0	S8 same heat	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:11
S13	254	S9 and heat	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:12

S14	2	S9 same (heat OR hsp\$)	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:12
S15	483	S7 with (\$virus OR \$viral)	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:16
S16	2	S15 same (heat\$6 OR hsp\$)	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:16
S17	544	S7 same (\$virus OR \$viral)	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:16
S18	2	S17 same (heat\$6 OR hsp\$)	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:17
S19	8262	(heat\$6 OR hsp\$) near2 promoter	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:17
S20	4217	S19 with (\$virus OR \$viral)	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:17
S21	1	S20 with essential	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:19
S22	417	S20 with gene	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:20
S23	16	S20 with replicat\$	US-PGPUB; USPAT; EPO; JPO; DERWENT	OR	ON	2005/04/12 17:20